510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
ASSAY AND INSTRUMENT COMBINATION TEMPLATE

A. 510(k) Number:

k053597

B. Purpose for Submission:

New device

C. Measurand:

B-type natriuretic peptide

D. Type of Test:

Quantitative

E. Applicant:

i-STAT Corporation

F. Proprietary and Established Names:

i-STAT BNP test

i-STAT Control Level 1

i-STAT Control Level 2

i-STAT Control Level 3

i-STAT BNP Calibration Verification Control Set

G. Regulatory Information:

1. Regulation section:
   862.1117, B-type natriuretic peptide test system

   862.1660, Single (specified) analyte controls (assayed and unassayed)

2. Classification:

   Class II, Class I

3. Product code:

   NBC, JJX

4. Panel:

   75 Chemistry

H. Intended Use:

1. Intended use(s):

   The i-STAT BNP test is an in vitro diagnostic test for the quantitative measurement of B-type natriuretic peptide (BNP) in whole blood or plasma samples using EDTA as the anticoagulant. BNP measurements can be used as an aid in the diagnosis and assessment of severity of congestive heart failure.

   The i-STAT Controls are assayed liquid plasma used to verify the integrity of newly received i-STAT BNP cartridges.

   The i-STAT BNP Calibration Verification Controls are assayed liquid plasma used to verify the calibration of i-STAT BNP cartridges throughout the reportable range.

2. Indication(s) for use:

   See Intended use(s) above.

3. Special conditions for use statement(s):

   Prescription use only

4. Special instrument requirements:

   i-STAT 1 Analyzer

I. Device Description:

Each i-STAT BNP cartridge provides a sample inlet, sensors to detect the BNP, and all the necessary reagents needed to perform the test. The cartridge contains a buffer and preservatives. A list of reactive ingredients is indicated below:

Reactive Ingredient	Biological Source
Antibody/Alkaline Phosphatase Conjugate	Murine IgG:Bovine Intestine
IgG	Caprine IgG: Murine IgG
Sodium Aminophenyl Phosphate	N/A
Heparin	Porcine Intestine

The i-STAT BNP Controls are supplied as assayed frozen liquid plasma at 3 levels, Control Level 1, Control Level 2 and Control Level 3. The human sera used in the preparation of this product has been tested by FDA approved test methods and found negative/non-reactive for HIV-1, HIV-2, HBsAg, HCV, HTLV-1 and HTLV-2.

The i-STAT Verification Control Set is supplied as 3 levels of assayed frozen liquid plasma at 3 levels, Level 1, Level 2 and Level 3. The human sera used in the preparation of this product has been tested by FDA approved test methods and found negative/non-reactive for HIV-1, HIV-2, HBsAg, HCV, HTLV-1 and HTLV-2.

J. Substantial Equivalence Information:

1. Predicate device name(s):

   Biosite Triage BNP Test

2. Predicate 510(k) number(s):

   k021317

3. Comparison with predicate:

Similarities		
Item	Device	Predicate
Assay methodology	Two-site ELISA	Two-site ELISA
Capture site	Heterogeneous	Heterogeneous
Capture antibodies	Monoclonal	Monoclonal
Enzyme label antibody	Monoclonal	Monoclonal
Sample type	Whole blood or plasma	Whole blood or plasma
Acceptable samples	EDTA anti-coagulated blood or plasma	EDTA anti-coagulated blood or plasma

Differences		
Item	Device	Predicate
Enzyme label	Fluorescent dye	Alkaline phosphatase
Enzyme detection	Fluorescent	Electrochemical
Sample volume	250 µL	20 µL
Reportable range	15-5000 pg/mL	5-5000 pg/mL

K. Standard/Guidance Document Referenced (if applicable):

CLSI Guideline EP7-A; CLSI Guideline EP9-A2; CLSI Guideline C-28-A2

L. Test Principle:

The i-STAT BNP test cartridge uses a two-site enzyme-linked immunosorbant assay (ELISA) method. Antibodies specific for BNP are located on an electrochemical sensor fabricated on a silicon chip. Also deposited in another location on the sensor silicon chip is an antibody/alkaline phosphatase enzyme conjugate specific to a separate portion of the BNP molecule. The whole blood or plasma sample is brought into contact with the sensors allowing the enzyme conjugate to dissolve into the sample. The BNP within the sample becomes labeled with alkaline phosphatase and is captured onto the surface of the electrochemical sensor during an incubation period of approximately seven minutes. The sample is washed off the sensors, as well as excess enzyme conjugate. Within the wash fluid is a substrate for the alkaline phosphatase enzyme. The enzyme bound to the antibody/antigen/antibody sandwich cleaves the substrate releasing an electrochemically detectable product. The electrochemical (amperometric) sensor measures this enzyme product which is proportional to the concentration of BNP within the sample.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

a. Precision/Reproducibility:

Precision data were collected as follows: duplicates of each control were tested daily for a period of 20 days for each of 3 lots of cartridges, resulting in a total of 434 replicates. The average statistics are presented below.

Aqueous Control	Mean	% CV (within-run)	% CV (total)
Level 1	126	9.0	11.1
Level 2	1551	6.6	8.1
Level 3	3337	8.0	9.8

Whole blood imprecision data were collected as follows: whole blood samples from 5 healthy donors were spiked to low, intermediate and high BNP concentrations affording 15 samples, each of which was measured in 10 i-STAT BNP cartridges from a single cartridge lot; three lots of cartridges were employed. The mean within-sample BNP concentration ranged from 84 – 3925 pg/mL and the within-sample imprecision (%CV) ranged from 3.4 to 9.4%; the average BNP concentration and imprecision were 1464 pg/mL and 6.5% respectively. The individual results are presented in the table below:

Donor	Mean BNP in pg/mL	% CV
1	99	6.3
1	765	3.4

Donor	Mean BNP in pg/mL	% CV
1	3803	4.1
2	107	7.8
2	1049	7.8
2	2638	5.7
3	108	9.2
3	1036	6.5
3	3805	5.4
4	84	9.1
4	783	9.4
4	3925	7.6
5	95	7.0
5	763	3.9
5	2900	4.8

b. Linearity/assay reportable range:

The dilution linearity of the i-STAT BNP test was studied using EDTA whole blood and plasma samples derived from 3 separate donors. For each donor, the original BNP negative sample and a BNP spiked sample were prepared. This process yielded three BNP positive whole blood samples that were then assayed in duplicate for each of 3 separate i-STAT BNP cartridge lots. These whole blood samples were then diluted using an equal mass of the original unspiked whole blood and assayed in duplicate. From this whole blood data, the BNP recovery was calculated.

Whole blood	**Concentration**	**Diluted concentration**	**% recovery**
A	590	312	106%
B	2765	1429	103%
C	5123	2803	109%

The plasma derived from these three donors was combined in all pair-wise combinations in equal volumes. These combinations were then assayed in duplicate for each of 3 separate i-STAT BNP cartridge lots. The BNP recovery for each pair was calculated using the average of the 6 results.

Plasma Blood Sample	Concentration pg/mL)	Diluted Concentration (pg/mL)	% Recovery
A	590	—	—
B	2764	—	—
C	5123	—	—
A+B	—	1570	94%
B+C	—	3992	101%
A+C	—	2734	96%

A plasma sample was spiked with BNP to a value of approximately 5000 pg/mL. This sample was subjected to a series of dilutions with fresh, un-spiked plasma in order to prepare a range of concentrations. The concentration of each sample/dilution was calculated based on the measured concentration of the initial solution and the dilutions performed. The diluted samples were then measured in i-STAT BNP test cartridges (N = 6-10). The procedure was repeated with a whole blood sample. The results of these experiments are summarized in the following table:

Sample	Dilution	Calculated [BNP] (pg/mL)	Measured [BNP] (pg/mL)	%Recovery
Plasma	1	52	57	110%
Plasma	2	104	114	110%
Plasma	3	259	265	103%
Plasma	4	518	560	108%
Plasma	5	1036	1002	97%
Plasma	6	2072	2277	110%
Plasma	7	3107	3384	109%
Plasma	8	4143	4222	102%
Whole Blood	1	44	41	93%
Whole Blood	2	88	88	100%
Whole Blood	3	269	287	107%
Whole Blood	4	537	554	103%
Whole Blood	5	725	720	99%
Whole Blood	6	1450	1367	94%
Whole Blood	7	3042	2826	93%
Whole Blood	8	4056	3856	95%

c. *Traceability, Stability, Expected values (controls, calibrators, or methods):*

The i-STAT BNP calibrators are traceable to an internal reference standard that has been prepared gravimetrically with synthetic BNP. The internal reference standard underwent a one-time value assignment to align with the ARCHITECT BNP assay with a decision threshold of 100 pg/mL. Manufacturers working calibrators are prepared by gravimetric manipulation of the standard and incorporate a one-time value assignment for alignment of methods. The i–STAT, AxSYM and ARCHITECT assays have been designed, by virtue of their calibration, to report comparable values. The i-STAT vs. ARCHITECT method comparison data exhibits a correlation slope of 0.97 (see method comparison section). Similar data for the ARCHITECT vs. AxSYM exhibited a slope of 1.03.

Stability studies were performed to evaluate the intended storage (open and closed vial) for the i-STAT controls and calibration verification materials. The real-time frozen stability of BNP control/calibration verification materials was established for 3 lots of material, each comprised of 3 levels. Stability was judged to be acceptable provided that the mean BNP concentration measured at each test event be within ± 20 % of the original mean concentration. The stability was acceptable over 5 months frozen storage. The stability studies are ongoing.

The labeling for the i-STAT controls and calibration verification materials states that, after thawing, the opened or unopened vial is stable for 4 hours when capped and stored at 2 – 8 º. Stability studies performed support the 4 hour time limit.

d. Detection limit:

The limit of the blank for the BNP method is 15 pg/mL, which is the lowest BNP level that can be distinguished from zero. The value was estimated using a control material with < 5 pg/mL BNP during a 20 day precision study in which 3 separate lots of BNP test cartridges were tested in duplicate using a pool of 6 i-STAT 1 analyzers for a total of 147 test results.

e. Analytical specificity:

The following muscle proteins were tested at both 1000 pg/mL and 20,000 pg/mL concentrations and found to have no detectable cross-reactivity for BNP: ANP, CNP, and N-terminal pro-BNP.

The i-STAT BNP assay employs electrochemical rather than optical detection. An electrogenic substrate is cleaved by an enzyme label giving rise to an electroactive product that can be oxidized at a sensor electrode generating a signal comprised of electrical current, therefore optical interferents, including hemoglobin, bilirubin, and chylomicrons, do not interfere with this mode of detection.

The following substances were found to have no significant effect (less than 10%) on the BNP method, when added to a plasma pool containing approximately 1000 pg/mL of B-type natriuretic peptide at the concentrations indicated:

Compound	Test Level (µmol/L unless otherwise indicated)
Acetaminophen	1660
Allopurinol	294
Ampicillin	152
Ascorbic Acid	227
Acetyl Salicylic Acid	3333
Atenolol	37.6
Caffeine	308
Captopril	23
Chloramphenicol	155
Diclofenac	169
Digoxin	6.15
Dopamine	5.87
Enalaprilat	0.86
Erythromycin	81.6
Furosemide	181
Sodium Heparin	90 U/mL
Ibuprofen	2425
Isosorbide dinitrate	636
Methyldopa	71
Nicotine	6.2
Nifedipine	1.156
Phenytoin	198
Propanolol	7.71
Salicylic Acid	4.34
Theophylline	222
Verapamil	4.4
Warfarin	64.9

f. Assay cut-off:

BNP results less than or equal to 100 pg/mL are representative of normal values in patients without CHF. See Clinical cut-off section below.

2. Comparison studies:

a. Method comparison with predicate device:

Method comparison data were collected using CLSI guideline EP9-A2. Venous blood samples were collected in EDTA evacuated tubes and analyzed in duplicate on the i-STAT System. A portion of the specimen was centrifuged and the separated plasma was analyzed in duplicate on the i-STAT 1 System and on the comparative method, the Abbott ARCHITECT BNP assay, within 1 hour of collection. Deming regression analysis was performed on the first replicate of each sample. In the method comparison table, n is the number of specimens in the first data set, Sxx and Syy refer to estimates of imprecision based on the duplicates of the comparative and the i-STAT methods respectively. Sy.x is the standard error of the estimate, and r is the correlation coefficient. The samples had BNP values ranging from 5-5000 mg/dL.

Method Comparison

	Abbott ARCHITECT
N	433
Mean (pg/mL)	482.1
Sxx (pg/mL)	38.1
Syy (pg/mL)	97.6
Slope	0.971
Intercept	-14.4
Sy.x	198.0
Xmin	5
Xmax	4797.7
Correlation, r	0.961

b. Matrix comparison:

EDTA plasma is the only sample type indicated. The labeling states that performance characteristics have not been established for samples taken from capillary tubes and direct skin punctures (e.g. fingersticks) so these sample types should not be used with the BNP cartridge.

3. Clinical studies:

Clinical studies performed with the Abbott AxSYM BNP assay are included in the labeling for the i-STAT BNP assay. The applicant provided the following to support the transfer of reference ranges:

- The AxSYM, ARCHITECT and i-STAT BNP assays employ an identical antibody set. The average imprecision is similar for the 3 assays as follows: AxSYM average %CV = 7.9 %; ARCHITECT average %CV = 5.2 %; i-STAT average %CV = 9.7 %.
- The i–STAT, AxSYM and ARCHITECT assays have been designed, by virtue of their calibration, to report comparable values.
- The CLSI document C28-A2, How to Define and Determine Reference Intervals in the Clinical Laboratory, provides guidance concerning the transferability of reference ranges from one measurement system to another. The i-STAT vs. ARCHITECT method comparison data exhibits a correlation slope of 0.97 (see method comparison section above). Also, similar data for the ARCHITECT vs. AxSYM exhibited a slope of 1.03 (see k060964).

a. Clinical Sensitivity:

In studies performed with the AxSYM BNP Assay, age-matched analysis of the heart failure and non-heart failure populations was performed based on the data published by the American Heart Association in the 2000 Heart and Stroke Statistical Update and according to the age structure of the United States population. The age distributions in the intended use population are approximately as follows: individuals less than 45 years old comprise 9%, individuals 45-54 years old comprise 11%, individuals 55-64 years old comprise 22%, individuals 65-74 years old comprise 26%, and individuals 75 years and older comprise 32%. The resulting combined AUC is 0.87 (0.85 to 0.90, 95%CI). The clinical sensitivity and specificity using a decision threshold of 100 pg/mL is presented in the table below.

	Males (Age Group)					
	All	<45 Years	45-54 Years	55-64 Years	65-74 Years	75+ Years
Sensitivity	71.0% (328/462)	47.1% (8/17)	57.1% (24/42)	57.3% (51/89)	70.6% (115/163)	86.1% (130/151)
95% Confidence Interval	66.6 to 75.1%	23.0 to 72.2%	41.0 to 72.3%	46.4 to 67.7%	62.9 to 77.4%	79.5 to 91.2%
Specificity	94.8% (403/425)	97.2% (104/107)	100.0% (71/71)	97.9% (92/94)	88.7% (102/115)	89.5% (34/38)
95% Confidence Interval	92.3 to 96.7%	92.0 to 99.4%	94.9 to 100.0%	92.5 to 99.7%	81.5 to 93.8%	75.2 to 97.1%

	Females (Age Group)					
	All	<45 Years	45-54 Years	55-64 Years	65-74 Years	75+ Years
Sensitivity	80.5% (186/231)	44.4% (4/9)	73.3% (11/15)	50.0% (13/26)	80.6% (58/72)	91.7% (100/109)
95% Confidence Interval	74.8 to 85.4%	13.7 to 78.8%	44.9 to 92.2%	29.9 to 70.1%	69.5 to 88.9%	84.9 to 96.2%
Specificity	88.4% (411/465)	95.9% (94/98)	90.7% (68/75)	89.6% (69/77)	85.7% (114/133)	80.5% (66/82)
95% Confidence Interval	85.1 to 91.2%	89.9 to 98.9%	81.7 to 96.2%	80.6 to 95.4%	78.6 to 91.2%	70.3 to 88.4%

b. Clinical specificity:

See Clinical Sensitivity section above.

c. Other clinical supportive data (when a. and b. are not applicable):

4. Clinical cut-off**:**

Data from the clinical studies performed with the AxSYM BNP assay were used to generate The Receiver Operating Characteristic (ROC) curve of BNP decision thresholds versus clinical sensitivity and clinical specificity. At a decision threshold of 100 pg/mL, the BNP assay demonstrated a clinical sensitivity and specificity of 74.2% and 91.5% respectively. The area under the curve is 0.90 (0.86 to 0.92, 95% CI).

5. Expected values/Reference range:

Plasma samples from 890 individuals (465 females, 425 males) who had not been diagnosed with heart failure were tested with the AxSYM BNP assay. This population included non-hospitalized patients with renal disease (not on dialysis), diabetes, hypertension and chronic obstructive pulmonary disease. BNP levels for these patients were not statistically different from the population of apparently healthy individuals. The data are summarized below.

Non-Heart Failure Population - All (Age Group)

	All	<45 Years	45-54 Years	55-64 Years	65-74 Years	75+ Years
Sample Size (N=)	890	205	146	171	248	120
Median (pg/mL)	21	17	9	24	23	31
Mean (pg/mL)	39	28	21	37	47	63
SD (pg/mL)	66	36	30	48	80	109
95th Percentile	135	85	87	119	160	254
Percentage < 100 pg/mL	91.5%	96.6%	95.2%	94.2%	87.1%	83.3%
Minimum (pg/mL)	0	0	0	0	0	0
Maximum (pg/mL)	907	263	142	380	907	837

Non-Heart Failure Population - Males (Age Group)

	All	<45 Years	45-54 Years	55-64 Years	65-74 Years	75+ Years
Sample Size (N=)	425	107	71	94	115	38
Median (pg/mL)	14	12	1	17	21	37
Mean (pg/mL)	30	23	9	26	47	49
SD (pg/mL)	61	34	14	45	96	51
95th Percentile	104	73	40	80	150	121
Percentage < 100 pg/mL	94.8%	97.2%	100.0%	97.9%	88.7%	89.5%
Minimum (pg/mL)	0	0	0	0	0	0
Maximum (pg/mL)	907	200	57	380	907	254

Non-Heart Failure Population - Females (Age Group)

	All	<45 Years	45-54 Years	55-64 Years	65-74 Years	75+ Years
Sample Size (N=)	465	98	75	77	133	82
Median (pg/mL)	26	23	23	37	23	25
Mean (pg/mL)	46	34	34	51	46	69
SD (pg/mL)	70	37	36	48	63	126
95th Percentile	150	89	111	155	159	266
Percentage < 100 pg/mL	88.4%	95.9%	90.7%	89.6%	85.7%	80.5%
Minimum (pg/mL)	0	0	0	0	0	0
Maximum (pg/mL)	837	263	142	230	374	837

Plasma samples from 693 patients with diagnosed heart failure (231 females, 462 males) were tested with the AxSYM BNP assay. All patients in this population were categorized according to the functional classification system published by the New York Heart Association (NYHA). This system divides heart failure patients into one of four categories of increasing disease progression (classes I to IV) based upon a subjective assessment of the patient's clinical signs and symptoms. The data from this study are summarized below.

Heart Failure Population – All

		NYHA Functional Class			
	All	I	II	III	IV
Sample Size (N=)	693	124	319	190	60
Median (pg/mL)	298	133	266	335	1531
Mean (pg/mL)	578	320	432	656	1635
SD (pg/mL)	771	388	574	841	1097
5th Percentile	14	9	15	12	188
95th Percentile	2154	1257	1534	2516	>4000
Percentage ≥ 100 pg/mL	74.2%	58.1%	73.0%	79.0%	98.3%
Minimum (pg/mL)	0	3	0	0	14
Maximum (pg/mL)	>4000	1651	>4000	>4000	>4000

Heart Failure Population – Males

	NYHA Functional Class				
	All	I	II	III	IV
Sample Size (N=)	462	94	215	121	32
Median (pg/mL)	268	122	258	293	1645
Mean (pg/mL)	524	314	409	597	1646
SD (pg/mL)	719	390	539	821	1032
5th Percentile	12	9	14	22	265
95th Percentile	1976	1281	1356	2288	3654
Percentage ≥ 100 pg/mL	71.0%	56.4%	70.7%	76.0%	96.9%
Minimum (pg/mL)	0	3	0	0	14
Maximum (pg/mL)	>4000	1408	3782	>4000	>4000

Heart Failure Population - Females

	NYHA Functional Class				
	All	I	II	III	IV
Sample Size (N=)	231	30	104	69	28
Median (pg/mL)	385	174	298	466	1408
Mean (pg/mL)	685	341	481	760	1623
SD (pg/mL)	858	388	641	870	1186
5th Percentile	16	14	21	12	244
95th Percentile	2593	1022	2031	2718	>4000
Percentage ≥ 100 pg/mL	80.5%	63.3%	77.9%	84.1%	100.0%
Minimum (pg/mL)	0	10	0	0	173
Maximum (pg/mL)	>4000	1651	>4000	>4000	>4000

N. Instrument Name:

i-STAT 1 Analyzer

O. System Descriptions:

1. Modes of Operation:

Single use cartridge

2. Software:

   FDA has reviewed applicant's Hazard Analysis and software development processes for this line of product types:

   Yes ___X_____ or No _____

3. Specimen Identification:

   Bar code reader is incorporated into the system

4. Specimen Sampling and Handling:

   Whole blood samples are applied directly into the sample well of the cartridge

5. Calibration:

   Factory set

6. Quality Control:

   The reliability of the results is maintained through a combination of user testing and instrument self-checks. The self checks occur with every cartridge run and verify performance of the analyzer and cartridge sub-systems. This includes checks on the individual sensor's performance, the integrity of the calibrant fluid, the response of the pressure and thermal transducers, and the flow of calibrant and sample within the cartridge. Any values that are statistically deviant from the factory established expectation values would cause the test results to be suppressed. Daily monitoring is through the use of internal and external electronic simulators. Liquid controls are provided for the verification of cartridge lot performance for all newly received cartridge lots.

P. Other Supportive Instrument Performance Characteristics Data Not Covered In The "Performance Characteristics" Section above:

Q. Proposed Labeling:

The labeling is sufficient and it satisfies the requirements of 21 CFR Part 809.10.

R. Conclusion:

The submitted information in this premarket notification is complete and supports a substantial equivalence decision.